Final Technical Report
EPA Assistance Agreement X-974569020-TNN
Project Title: Study of Honey Bee Colonies (To assist the University in implementing a program to reduce chemical acaricide use in honey bee colonies and prevent hive product contamination)

James Patrick Parkman
Department of Entomology and Plant Pathology
University of Tennessee
Knoxville, TN 37996-4560
(865) 974-7135
jparkman@utk.edu

The *Varroa* mite, *Varroa destructor*, is the most damaging pest to honey bees in the US and much of the world. But only two chemical acaricides (treatments for mite control) are available in the US for *Varroa*: Apistan™ (contains fluvalinate, a pyrethroid insecticide) and CheckMite+™ (contains coumaphos, an organophosphate insecticide); and resistance to both of these products by *Varroa* is becoming widespread. Because of resistance development and in response to beekeepers' desire for safer and more sustainable mite control, other mite management tools have been developed. The goal of this project was to provide Tennessee beekeepers with some of these tools, mite-resistant bee stock and open bottom boards (which exclude mites from the colony after they fall from bees); and to demonstrate their correct use, in hopes of reducing chemical acaricide use.

Methodology

An application to participate was provided to members of the Tennessee Beekeepers Association (TBA) via the Association's newsletter in October and November 2002. Ten beekeepers from across the state were chosen from the pool of applicants. Participating beekeepers and their apiaries (bee yards) were located in Knox, Loudon, Sevier and Union counties in east Tennessee; Bedford, Montgomery, Moore, Robertson and Williamson counties in central Tennessee; and Shelby County in west Tennessee.

Each beekeeper was to provide 20 bee colonies. Ten were to be requeened with *Varroa*-resistant queens and the bottom boards were to be replaced with open bottom boards. (These colonies are hereafter referred to as *Varroa*-managed.) Ten were to be requeened with mite-susceptible queens and continue to be maintained over closed bottom boards. (These colonies are hereafter referred to as non-managed.) Beekeepers were to monitor mite population levels once every 4 weeks, using bottom board sticky traps provided by the project, until late September 2003.

Queens, open bottom boards and sticky traps were delivered by University project personnel to all participants from mid-April to mid-May 2003. Beekeepers were assisted in requeening bee colonies and replacing closed bottom boards with open bottom boards; and shown how to monitor for mites using sticky traps. They were provided data sheets, for recording mite numbers, and treatments (Apistan™ and CheckMite+™ for non-managed colonies; and ApiLife VAR™ and Apiguard™, both containing plant essential oil-based active ingredients, for *Varroa*-managed colonies) to be used if mite populations reached damaging

levels (based on trap collections). The damaging level, referred to as a treatment threshold, has been determined (using our design of sticky trap) to be 25 mites collected per 24 hours for an averaged-sized bee colony.

If treatments were applied, beekeepers were asked to collect wax and honey samples from treated colonies after treatments were removed. Samples were analyzed for treatment residues in the lab using gas chromatography. Samples were to be taken only from frames (supplied by project personnel) that held undrawn wax foundation. These frames could only contain wax and honey produced during the current (2003) season; thus, any residues found in samples taken from these frames could have originated only from treatments made during that season.

To better quantify *Varroa* management practices and adoption of non-chemical tools, a questionnaire was circulated to TBA members in fall of 2003. Beekeepers were questioned about their use of mite-resistant bee stock and open bottom boards in 2002 and 2003.

Educational materials, a printed publication and a slide presentation, were developed to promote sustainable management of *Varroa*. These materials were developed for use primarily by University of Tennessee Agricultural Extension agents and the state's beekeepers.

Results and Discussion

The beekeeping community is receptive to new ideas and technology for managing pests. This is evidenced by the eager participation of Tennessee's beekeepers and the TBA in this demonstration project. Adoption of non-chemical *Varroa* management tools and techniques is increasing in Tennessee (see results of the TBA questionnaire) because the University Apiculture Program has been promoting their use, not only through this project, but also through presentations made over several years to beekeeping organizations throughout the state. Undoubtedly, adoption has increased also because more management tools have become commercially available in the last few years.

One result of the project, however, illustrated a problem (hopefully, temporary) in obtaining effective management tools. Most queens purchased from commercial queen producers and provided to project participants did not exhibit resistance based on sticky board sampling results (see "*Varroa* monitoring" section below). Lack of resistance was probably a result of suboptimal queen mating conditions. Weather conditions in northern California and the southeastern US (locations of the queen producers from whom we purchased) during much of late winter/early spring 2003 were cool and wet, and, thus, not conducive to successful queen-mating. Also, producers may not yet have perfected conditions to insure resistant queens are mating with resistant drones. Open-mating (the mating of a virgin queen with any drones encountered on her mating flight) is standard procedure for commercial queen production. However, for an open-mated, resistant queen to produce predominantly resistant worker progeny, she must mate with resistant drones. Mating with resistant drones, and preservation of resistant traits, could be facilitated by improved isolation of mating yards and by saturating drone congregation areas near mating yards with resistant drones. (FYI: Mothers of resistant queens and drones are artificially inseminated by queen breeders with sperm from resistant drones to insure these "breeder" queens produce only resistant offspring.)

Below are detailed results for project activities including management effects on V*arroa* populations, treatment residue analysis, management questionnaire, and development and dissemination of educational materials.

***Varroa* monitoring.** *Varroa* population numbers increased, on average, 19-fold over the course of the project (April to September 2003) in *Varroa*-managed bee colonies. In non-managed colonies, *Varroa* populations increased 24-fold during the same time period. Although mite numbers increased steadily in most colonies, the treatment threshold was never reached in 29% of *Varroa*-managed colonies, and in 17% of non-managed colonies. Eighteen percent of *Varroa*-managed colonies died during the project, 19% of non-managed colonies died. (Only colonies surviving for the duration of the project were used to determine results of *Varroa* sampling.) Cause of death for colonies was not determined. Probable causes were *Varroa* infestation and/or queen failure (suboptimal egg production caused by poor mating conditions).

Residue analysis. Eighteen honey or wax samples were collected from acaricide-treated colonies by participants. Seven wax and three honey samples were taken from Apiguard™-treated colonies; four wax and two honey samples were taken from ApiLife VAR™-treated colonies. These samples were analyzed for thymol residues, the principal active ingredient in these two products. A honey sample was each taken from an Apistan™-treated and a CheckMite+™-treated colony and were analyzed for fluvalinate and coumaphos, respectively.

One wax sample each taken from an Apiguard™- and an ApiLife VAR™-treated colony contained notable amounts of thymol residues. The wax sample from the ApiLife VAR™-treated colony contained 9.7 ppm thymol; the sample from the Apiguard™-treated colony contained 103 ppm. Thymol is detectable in honey by taste at levels between 1.1 and 1.6 ppm; however, the relationship between residues in wax and resulting residues in honey stored in that wax has not been adequately studied. Because the application of these products was not supervised by project personnel, wax contamination may have resulted from product misuse. Exemptions from the requirement of tolerance has been established by EPA for residues of thymol in honey and beeswax. Apiguard™ is an experimental product but contains about the same amount of thymol as ApiLife VAR™. If these products are applied properly, residues of thymol should not accumulate in honey or wax to levels found here.

No residues of fluvalinate or coumaphos were detected in the two honey samples taken from colonies treated with Apistan™ and CheckMite+™, respectively.

The number of samples taken does not necessarily reflect the number of treatments applied. Participants were required to take samples only from the new frames we provided. There may not have been any wax and honey on these frames from which to take samples. Because of persistent wet weather, 2003 was a below-average year for honey production in Tennessee (and, consequentially, wax production because honey is converted to wax by worker bees) and this undoubtedly affected the number of samples submitted. Many participants reported that the new frames provided had not been “drawn” (had beeswax comb constructed on them) by worker bees.

Questionnaire results. Forty-two beekeepers, with honey bee colonies in 43 counties, responded to our questionnaire. Average number of colonies owned per responding beekeeper in 2002 was 27; in 2003 the average number was 30. Twenty-nine percent of respondents used *Varroa*-resistant queens in 2002. Use of resistant stock among respondents increased to 41% in 2003. Twenty-three percent believed that their resistant queens provided poor mite control; however, 23% thought that their queens provided fair control. Eighteen and 5% believed they received good and excellent control, respectively, with their resistant queens.

Open bottom board use is apparently more common. In 2002, 62% of beekeepers used open bottom boards; 76% used open bottom boards in 2003. Only 3% thought open bottom boards provided poor control. Sixteen, 50 and 9% believed their open bottom boards provided fair, good and excellent *Varroa* control, respectively.

Educational Materials. An Extension publication, "Managing *Varroa* Mites in Tennessee," was written and posted on the Publications section of the University's Apiculture Program website (http://eppserver.ag.utk.edu/Bees/test/Intro.html). An accompanying PowerPoint presentation was also produced and posted on the Apiculture website. The publication and presentation are also being made available on CD to each county Extension office in Tennessee. County Extension agents will be able to make the presentation to stakeholders in their county. Subject matter for the publication and presentation includes *Varroa* biology and damage symptoms, monitoring (sampling), treatment options, and non-chemical management tools and tactics.

Beekeeping in the US changed dramatically with the introduction and spread of *Varroa*; and the development of resistance by the pest to chemical acaricides used for its control has only worsened the beekeeper's plight. As educators, we are responsible for keeping beekeepers informed on the best *Varroa* management technology available. The funding provided through EPA's SAIG program has allowed us to promote and demonstrate sustainable *Varroa* management tools and strategies to beekeepers across Tennessee. Lack of mite resistance in queens used in the project was disappointing, but this result does not lessen the importance of non-chemical management of *Varroa*. We believe non-chemical tools and strategies, especially when used in combination, can provide long-term, sustainable management of *Varroa* and, thus, reduce dependence on traditional chemical acaricides and contamination of hive products.